Effect of somatostatin on growth hormone and prolactin response to dermorphin in man

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Abstract. The effects of iv somatostatin (somatotrophin release inhibiting factor (SRIF)) on growth hormone (GH) and prolactin (Prl) response to dermorphin (D) were tested in 6 healthy men. In all subjects D induced a significant increase in GH and Prl levels, as expected. SRIF completely blocked the GH-releasing activity of D, whereas it only reduced the Prl-releasing activity. The results confirm that D is a potent stimulant for GH and Prl release in man, and furthermore demonstrate that the action of D on GH secretion can be completely overridden by SRIF.

Recently we reported that dermorphin (D), a new potent opioid heptapeptide (H-Tyr-D-Ala-Phe-Gly-Tyr-Pro-Ser-NH₂) originally isolated from the skin of the South American frog (Broccardo et al. 1981) significantly increased plasma levels of growth hormone (GH) in man (degli Uberti et al. 1983a) and that this effect of D on GH release could be partially reduced by the opiate receptor antagonist naloxone, but not abolished.

Our finding is in accord with the large body of evidence which clearly shows that opioid peptides are capable of inducing the release of GH both in experimental animals (Chihara et al. 1978; Meites et al. 1979a,b; Rivier et al. 1977) and in man (Stubbs et al. 1978; von Graffenreid et al. 1978). Although the mechanism of such a release still remains to be clarified, it is generally accepted that opioid peptides do not exert their GH-releasing activity through a direct effect on the anterior pituitary gland, but may have a central mechanism of action, probably by modulating the secretion of hypothalamic releasing or release-inhibiting factor (Ferland et al. 1977; Imura et al. 1981; Koenig et al. 1980; Martin et al. 1975). To date there is no information concerning the mechanism and site of action of D in eliciting GH release.

It is well known that the GH secretion by the pituitary is controlled by a dual mechanism (Martin 1976) involving somatostatin (somatotrophin release inhibiting factor (SRIF)) (Brazeau et al. 1973) and GH-releasing hormone (Guillemin et al. 1982), and that SRIF is a hypothalamic regulatory tetradecapeptide which inhibits GH release by the anterior pituitary.

In the present study we investigated the effect of SRIF administration on D-induced GH release in man. In addition, since D iv infusion stimulates the release of prolactin (Prl) in man (degli Uberti et al. 1983b) we have in the same study also evaluated the effects of SRIF on D-induced Prl release.

Materials and Methods

Six healthy men aged 19–32 years gave their informed consent to participate in this study. All subjects were within ± 10% of the ideal body weight and none of them took drugs or medicines. Each subject was admitted to

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the Clinical Centre at least 3 days before the study and tested on 3 separate occasions in random sequence with a 4 day interval between the tests.

The subjects fasted overnight and were supine until completion of each test. Forty min before test, a 19-gauge butterfly needle was inserted in an arm vein for blood sampling and a double lumen indwelling catheter was placed in a vein in the contralateral arm for the infusions (saline, D or SRIF). Patency was maintained by a slow iv drip of 0.9% saline. At 08.30 h (−30 min) an iv bolus of SRIF (250 µg) in 10 ml 0.9% saline was slowly injected followed by a constant iv infusion of SRIF in 40 ml 0.9% saline at a rate of 500 µg/h over 180 min (−30 to 150 min of the study). After 3 blood samples had been obtained (−30, −15 and 0 min), a 30 min infusion was started with D (5.5 µg/kg/min) in 30 ml 0.9% saline (SRIF + D study) or with 0.9% saline solution at 1 ml/min (SRIF + D-placebo study). The order of 30 min infusions (D or D-placebo) was alternated by a random sequence in a single blind schedule. Blood samples were then obtained 5, 15, 30, 45, 60, 90, 120, 150 and 180 min after the beginning of D or D-placebo infusions.

Fig. 1.

Mean ± SEM GH response to D (●) or D-placebo (○) in 6 normal males with either saline (—), or SRIF infusion (——). *P < 0.05; **P < 0.01; ***P < 0.005; ****P < 0.001 (vs mean basal value).
On another occasion the same subjects were studied with a protocol identical to that used in SRIF/D or SRIF/D-placebo studies with the following exception. At 08.30 h of the experimental day, instead of SRIF administration a bolus injection of 10 ml 0.9% saline was given iv, followed by a constant infusion of 0.9% saline at 0.15 ml/min over 180 min (−30 to 150 min of the study). After 3 blood samples had been obtained (−30, −15 and 0 min) a 30 min infusion was begun with D (5.5 µg/kg/ min) in 30 ml 0.9% saline.

Analytic procedures

Blood was drawn into vacutainer tubes containing EDTA-2Na and immediately centrifuged. All plasma samples were frozen within 10 min of collection, and stored at −30°C until assayed. Plasma Prl and GH levels were measured by a specific RIA using Biodata kits (Milan, Italy). The sensitivity of the assay was 1 and 0.15 ng/ml, respectively. The intra- and inter-assay coefficients of variation were, respectively: Prl 4.5 and 6.2%, and GH, 6.8 and 11.1%.

D was synthesized by a conventional method in solution as previously described (Salvadori et al. 1982), dissolved in sterile 0.15 m sodium chloride, and passed over a 0.45 µm millipore filter. D was available in lyophilised form and dissolved in 0.9% saline before iv administration. Somatostatin (Stilamin) was obtained from Serono (Milan, Italy).

Data analysis

Statistical analysis of data was performed using double-tailed Student’s t-test for paired and unpaired data and analysis of variance as applicable.

Results

As shown in Fig. 1, after D administration GH rose (D + saline), as expected, from a mean basal level (mean of the values observed at −30, −15 and 0 min) of 1.01 ± 0.5 ng/ml, to a peak of 29.09 ± 4 ng/ml at 60 min, decreasing slowly although remaining still significantly high 2 h after the end of D infusion. SRIF infusion lowered basal GH levels (SRIF + saline) slightly but not significantly. Following cessation of SRIF infusion, GH tended to rebound, reaching the mean level of 4.74 ± 1.22 ng/ml at 180 min, significantly (P < 0.05) higher than that observed prior to SRIF administration (0.99 ± 0.4 ng/ml).

The GH response to D was completely abolished (P < 0.001) by SRIF pre-treatment (SRIF + D). After the SRIF infusion was stopped, GH levels increased rapidly to a value of 12.54 ± 4.51 ng/ml significantly higher (P < 0.01) than that seen in basal pre-SRIF conditions (1.1 ± 0.46 ng/ml).

As shown in Fig. 2, D infusion (D + saline) produced the expected increase in plasma Prl from the mean basal level of 7.23 ± 1.76 ng/ml to a maximum of 40.75 ± 5.5 ng/ml (P < 0.001) at 45 min. SRIF infusion (SRIF + saline) had no significant effect on basal Prl levels. SRIF (SRIF + D) reduced the D-induced Prl release significantly (P < 0.05 at 30 and 45 min vs D + saline values).

No side effects were observed during SRIF infusion. A feeling of tingling in the whole body and heaviness in the legs were common side effects of D administration, as reported previously (degli Uberti et al. 1983a,b).

Discussion

These results confirm our previous reports that D is a potent stimulus for GH and Prl secretion in man (degli Uberti et al. 1983a,b). The present study has demonstrated that SRIF completely blocks the GH response to D. This finding is in agreement with the extensive studies showing that SRIF can inhibit not only the in vivo GH response to other stimuli such as insulin-induced hypoglycaemia, arginine, 1-dopa or exercise in man (Mortimer et al. 1974; Prange-Hansen et al. 1973; Rodriguez-Arnao et al. 1981; Siler et al. 1973) and pentobarbital or morphine in rats (Brazeau et al. 1974; Martin et al. 1975), but also in vitro GRF-stimulated release of GH by primary cultures of rat pituitary cells (Vale et al. 1983). However, in view of previous experimental evidence that SRIF over-rides all inducers of GH secretion via a direct action on the pituitary, our findings may only indicate that the inhibitory effect of SRIF on GH release cannot be overcome by the remarkable GH-releasing activity of D, and do not allow us to assume whether the stimulation of D on GH secretion is mediated via a direct action on the pituitary or through a hypothalamic mechanism.

In the control experiments, after the SRIF infusion was stopped, GH increased to levels significantly higher than those seen in basal conditions. This rebound has also been reported by Mortimer et al. (1974) and Rodriguez-Arnao et al. (1981), and is consistent with the concept of SRIF inhibitory influence on GH release. When D was infused with SRIF, the post-inhibitory rebound was more pro-
Mean ± SEM Prl response to D (●) or D-placebo (○) in 6 normal males with either saline (---), or SRIF infusion (----).

*P < 0.05; **P < 0.005; ***P < 0.001 (vs mean basal value).

This result might be accounted for by a longer GH-releasing activity of D in comparison with the brief duration of action of SRIF.

In this study, the additional result that SRIF, in doses that completely prevented the action of D on GH secretion, was capable of partially inhibiting the Prl response to D, is of interest, since a few reports have indicated that the inhibiting action of SRIF is not confined to GH and TSH, but is also exerted on Prl secretion. At present, however, results on the effect of SRIF on Prl release are conflicting (Carr et al. 1975; Gomez-Pan et al. 1979; Siler et al. 1974) and it is still not clear whether endogenous SRIF may participate in the regulation of Prl secretion (Besser et al. 1974; Copinschi et al. 1976; Vale et al. 1974; Yen et al. 1974; Rodriguez-Arnao et al. 1981). Therefore, in the absence of other data concerning the exact mechanism and site of action of D, any speculation does not seem reasonable in order to explain the
influence of SRIF on D-stimulated Prl secretion and establish which combination of factors may be involved in the apparently greater effect of SRIF in inhibiting the GH response rather than Prl response to D.

In conclusion, our findings confirm the remarkable potency of D in causing GH and Prl release, and demonstrate that SRIF can inhibit completely the GH response and partially Prl response to D. However, the precise mechanism and site of action of D still remain to be determined.

References


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